

OCCURRENCE OF BIOLOGIC FALSE POSITIVE REACTIONS WITH RPR (CIRCLE) CARD TEST ON LEPROSY PATIENTS

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PRECIPITATION TESTS used in the serodiagnosis of syphilis are known to be capable of showing biologic false positive reactions in persons with other diseases or conditions. Such reactions have been reported in persons with leprosy, malaria, respiratory infections, infectious mononucleosis, undulant fever, measles, and vaccinia as well as in pregnant women (1). The globulin responsible for the biologic false positive reaction differs from the syphilis antibody particularly because it is inhibited by a lecithin associated with the globulin fraction of human serum. Kahn considered the antibody to be the result of auto-immunization by lipids released during exaggerated tissue breakdown which occurs in the conditions mentioned, particularly malaria and leprosy (1).

The rapid plasma reagin (RPR) card test is a precipitation test for syphilis which was introduced by Portnoy and associates (2). A great deal of work has been devoted to the comparative control of this test and its modified version, the RPR (circle) card test, and a majority of the workers have emphasized its rapid performance, its simplicity, and its adequate sensitivity and specificity (2-9). In a recent study of the RPR card test for syphilis (10), several of us obtained data which fully agreed with the conclusions reported by these workers. However, to our knowledge, the RPR card test had never been studied on serums of patients

with leprosy, and biologic false positive reactions to the test had not been reported for this disease.

We performed the RPR (circle) card test in parallel with the Kahn standard reaction test on serums of leprosy patients who were under treatment at the Public Hospital for Infectious Diseases of Athens. On serums reactive by either or both of these tests, we also performed the fluorescent treponemal antibody (FTA) test. Our observations are reported here.

Materials and Methods

The serum samples used in this study were freshly collected from 50 leprosy patients after fasting; 39 of these patients had the lepromatous form of leprosy, six had the borderline (dimorphous) form, and five had the tuberculoid form. None of these donors had reported a history of syphilis.

Each serum sample was tested by the classic Kahn procedure after complement inactivation, using the cardioliipin of Behringwerke, A. G., Marburg, Germany, and by RPR (circle) card test, using the antigen of Hynson, Westcott & Dunning, Inc., Baltimore, Md. The antigen consisted of cardioliipin and carbon particles, choline chloride, and EDTA. The antigen- (unheated) serum reaction test was performed on plastic-coated cards containing 18-mm. circle spots. The cards were mechanically rotated for 8 minutes at 100 rpm. All serums positive by these tests were tested further by the specific FTA test (used as a standard test).

Results

As shown in table 1, 13 of the 39 patients with the lepromatous form of leprosy were reactive to the RPR (circle) card test and 12 were reactive to the Kahn test. None of the five patients

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with the tuberculoid form had a positive precipitation reaction and one of the six borderline patients was reactive.

Of the 50 serum samples examined, a total of 14 were reactive by the RPR (circle) card test and a total of 13 were reactive by the Kahn test. These reactive serums were then examined by fluorescence for specific treponemal antibodies. As shown in table 2, all were nonreactive by the specific FTA test.

Discussion and Conclusion

Certain features, not obviously the direct result of leprosy, have been observed in persons with this disease. For example, Bonomo and associates (11) reported positive tests for anti-thyroglobulin among leprosy patients, and Cathcart and associates (12) reported positive latex fixation tests for rheumatoid factor. Matthews and Trautman (13) described serum protein abnormalities such as a decrease of albumin and a rise of gamma, beta, and alpha 2 globulins.

The occurrence of biologic false positive reactions in precipitation tests for leprosy has been demonstrated by many workers (14) who emphasized that nonspecific reactions occur especially in the lepromatous form of the disease.

Ruge (15) reported 16 percent biologic false positive reactions in a series of leprosy patients, and he observed a decrease in such reactions after antileprotic treatment. Matthews and Trautman reported that after extraction of the cryoprotein fraction from leprosy patients' serums, which had been previously positive by the VDRL test, the serums became negative to the VDRL test (13).

Table 1. Results of RPR (circle) and Kahn tests on serums from leprosy patients, by clinical forms of their disease

Clinical form	Number of patients	Number nonreactive		Number reactive	
		RPR card	Kahn card	RPR card	Kahn card
Lepromatous.....	39	26	27	13	12
Tuberculoid.....	5	5	5	0	0
Borderline (dimorphous)...	6	5	5	1	1
Total.....	50	36	37	14	13

Table 2. Results of FTA test on serums reactive by the RPR (circle) card or Kahn tests, by patients' sex and clinical form of leprosy

Sex and clinical form	RPR card	Kahn	FTA
<i>Females</i>			
Lepromatous	Nonreactive	Reactive	Nonreactive
Lepromatous	Reactive	Reactive	Nonreactive
Lepromatous	Reactive	Nonreactive	Nonreactive
Lepromatous	Reactive	Reactive	Nonreactive
Lepromatous	Reactive	Reactive	Nonreactive
Lepromatous	Reactive	Reactive	Nonreactive
Borderline (dimorphous)	Reactive	Reactive	Nonreactive
<i>Males</i>			
Lepromatous	Reactive	Reactive	Nonreactive
Lepromatous	Reactive	Reactive	Nonreactive
Lepromatous	Reactive	Nonreactive	Nonreactive
Lepromatous	Reactive	Reactive	Nonreactive
Lepromatous	Reactive	Reactive	Nonreactive
Lepromatous	Nonreactive	Reactive	Nonreactive
Lepromatous	Reactive	Nonreactive	Nonreactive
Lepromatous	Reactive	Reactive	Nonreactive
Lepromatous	Reactive	Reactive	Nonreactive

In our series of leprosy patients, serums which were reactive by the RPR (circle) card and the Kahn tests were nonreactive by the specific FTA test. This result, combined with the absence of a history of syphilis in our patients, supports our conclusion that the positive results obtained with the RPR (circle) card and the Kahn tests are biologic false positive reactions. These reactions occurred primarily among the patients with the lepromatous form of leprosy.

Our observations indicate that although the RPR (circle) card test is recommended as a routine screening test it has the same disadvantage of other precipitation tests for syphilis in that it shows biologic false positive reactions in patients with leprosy.

REFERENCES

- (1) Wilson, G. S., and Miles, A. A.: Topley and Wilson's principles of bacteriology and immunity. Ed. 5. Arnolds, London, 1966.
- (2) Portnoy, J., Brewer, J. H., and Harris, A.: Rapid plasma reagin card test for syphilis and other treponematoses. Public Health Rep 77: 645-652, August 1962.
- (3) Portnoy, J.: Modifications of the rapid plasma reagin (RPR) card test for syphilis for use in

- large scale testing. *Amer J Clin Path* 40: 473-479 (1963).
- (4) Reed, E. L.: The rapid plasma reagin (circle) card test for syphilis as a routine screening procedure. *J Conf Public Health Lab Directors* 23: 96-103 (1965).
 - (5) Enbring, H., Matthes, M., and Schulze Icking-Konert, O.: Studies of the most satisfactory protection against the transfer of syphilis by blood transfusion (with special regard to the cardiolipin micro flocculation test (CMT) and the RPR card test). *BIOTEST-Mitteilungen* No. 23, 1966.
 - (6) Nolting, S., and Fegler, F.: Der Plasma-Reagin-Kartenschnelltest zur Syphilisdiagnose. *Mun-chen Med Wschr* 108: 845-847 (1966).
 - (7) Reed, E. L.: Further studies of the specificity of the RPR (circle) card test for syphilis as revealed by treponemal testing and clinical diagnosis. *J Conf Public Health Lab Directors* 24: 203 (1966).
 - (8) Reed, E. L.: The combined use of the RPR card and FTA-ABS tests in the serodiagnosis of syphilis. *J Conf Public Health Lab Directors* 26: 123-134 (1968).
 - (9) Reed, E. L.: FTA-ABS confirmation of nontreponemal tests. Presented at FTA-ABS test seminar co-sponsored by the Maryland State Department of Public Health, Bureau of Laboratories, and NCDC Venereal Diseases Research Laboratory, Oct. 22, 1968.
 - (10) Achimastos, A., Papadopoulos, G., and Tolis, G.: The RPR card test. A rapid precipitation test in the serodiagnosis of syphilis. *Hellenic Armed Forces Med Rev* 3: 325-331, September 1969.
 - (11) Bonomo, L., Dammacco, F., Pinto, L., and Barbieri, G.: Thyroglobin antibodies in leprosy. *Lancet* No. 7312: 807-809, Oct. 19, 1963.
 - (12) Cathcart, E. S., Williams, R. C., Jr., Ross, H., and Calkins, E.: The relationship of the latex fixation test to the clinical and serologic manifestations of leprosy. *Amer J Med* 31: 758-765, November 1961.
 - (13) Matthews, L. J., and Trautman, J. R.: Clinical and serological profiles in leprosy. *Lancet* No. 7419: 915-917, Nov. 6, 1965.
 - (14) Edmundson, W. F., Wolcott, R. R., Olansky, S., and Ross, H.: A clinico-serologic study of leprosy. Part I. Result of serologic test for syphilis including the *Treponema pallidum* immobilization test. *Int J Leprosy* 22: 440-449, October 1954.
 - (15) Ruge, H.: Luesreaktionen an Lepraseren. *Medizinische Welt* 48: 2620 (1966).

Tearsheet Requests

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Pest-Free U.S. Military Equipment From Vietnam

The Public Health Service and the Department of Agriculture have begun an advisory program for the Department of Defense in the quarantine processing of equipment being returned from Vietnam. Military units returning in the future will bring all equipment home after it has been inspected for pests of medical and agricultural importance.

The greatest public health concern is to prevent importation of human diseases. The Department of Agriculture is concerned with animal diseases and a number of beetles, termites, and other insects.

The Foreign Quarantine Program of the National Communicable Disease Center in Atlanta, Ga., and the Department of Agriculture will advise the Department of Defense on inspection procedures for the retrograde cargo and in training military personnel to assist in inspection and quarantine clearance of the equipment.

Dr. Charles R. Joyce of the U.S. Quarantine Station, Honolulu, was the first Public Health Service adviser to Gen. Creighton W. Abrams, Commander of the U.S. Military Assistance Command, Vietnam. The first Department of Agriculture adviser was Charles D. Nigro of the Plant Quarantine Division of New York City. U.S. customs officials already assigned to duty in Vietnam will also assist.

Teams of three public health advisers and three agricultural advisers are serving tours of duty in the program. There will be at least four staging areas in Vietnam to process materiel for shipment by sea with 10 or more subareas at airports or other locations. Inspection will cover every item to be shipped as well as the cargo vessels and aircraft in which the equipment will be returned to the United States. Inspection teams will take detailed precautions to prevent contamination during storage while awaiting shipment.